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Caloplaca tianshanensis (lichen-forming *Ascomycota*), a new species of subgenus *Pyrenodesmia* from China

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Abstract — *Caloplaca tianshanensis* is described as a species new to science. It has a crustose and areolate thallus of yellowish-brown color with conspicuous cracks, bearing dark brown to black apothecia. An analysis of ITS sequences supports the affinity of the new species to subgenus *Pyrenodesmia*.

Key words — *Teloschistaceae*, peltate areoles, zeorine, isthmus

Introduction

As presently circumscribed, the subgenus *Pyrenodesmia* (A. Massal.) Boistel of the lichen-forming genus *Caloplaca* Th. Fr. (*Teloschistaceae*) contains lichens characterized by brown or black apothecia, an epihymenium that is usually K– or K+ violaceous, and a thallus that is not yellow, orange or red unlike most other *Caloplaca* spp., and lacks the K+ red reaction of the parietin complex (Tretiach & Muggia 2006).

Forty-two species of the genus *Caloplaca* were reported from China (Wei 1991). Among them 9 species belong to the subgenus *Pyrenodesmia*: *C. chrysophora* Zahlbr., *C. cupreorufa* Zahlbr. and *C. cervina* Zahlbr. from Sichuan (Zahlbruckner 1930, 1932), *C. giraldii* Jatta from Shaanxi (Jatta 1902) and Sichuan (Zahlbruckner 1930, 1931), *C. ochrotropa* Zahlbr. from Yunnan (Zahlbruckner 1930, 1932), *C. plumbeoolivacea* H. Magn., *C. circumalbata* (Delile) Wunder from Inner Mongolia (Magnusson 1944, as *C. aegyptiaca* (Müll.

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Table 1. Lichen species and sequences used to generate the phylogenetic tree.

Species	GenBank #
Caloplaca albopruinosa (Arnold) H.Olivier	EF093577
	EF093578
C. albopustulata Khods. & S. Y. Kondr.	EU192150
C. alociza (A. Massal.) Mig.	EF090933
	EF090936
C. badioreagens Tretiach & Muggia	EF081039
	EF081040
C. cerina (Ehrh.Ex Hedw.) Th.Fr.	AF353958
C. chalybaea (Fr.) Müll.Arg.	AY313970
	AY313971
C. chlorina (Flot.) Sandst.	AF353959
C. concreticola Vondrák & Khodos.	EU192153
	EU192152
C. cretensis (Zahlbr.) Wunder	EF093579
C. erodens Tretiach et al.	EF090922
	EF090921
C. obscurella (J. Lahm) Th.Fr.	AY313976
	AY313977
C. peliophylla (Tuck.) Zahlbr.	AY313965
C. tianshanensis Xahidin, A. Abbas & J.C. Wei [a]	GU552277
C. transcaspica.	EU192156
C. variabilis (Pers.) Müll. Arg.	EF090926
	EF090925

Arg.) Stnr; Wunder 1974), *C. transcaspica* (Nyl.) Zahlbr. from Inner Mongolia (Magnusson 1944, as *C. paulsenii*), Gansu, Qinghai (Magnusson 1940, as *C. paulsenii*) and Xinjiang (Poelt & Hinteregger 1993)., and *C. alociza* (Massal.) Mig. from Jiangsu (Wu & Xiang 1981, as *C. agardhiana* (Flot.) Flag., 1981).

During a study of the lichen genus *Caloplaca* in China numerous samples were collected by the first two authors from the Xinjiang region. Some specimens belonging to *Pyrenodesmia* attracted our special attention and were examined in detail for morphology, anatomy, chemistry and molecular systematics. As a result, one of them, *C. tianshanensis*, is described here as new to science.

Material and methods

Material

The lichen material examined for morphology, anatomy, chemistry and molecular analyses was collected from Miaoergou on Mt. Nan-shan in the Tianshan mountain chain, Xinjiang region, in 2009.

Morphological observations

Observations and photographs were made with a dissecting microscope (Leica MZ 12), a Zeiss Axioplan compound microscope and an Axiocam digital camera with associated software. Squash mounts and hand sections were routinely examined using tap water as the mounting medium. Lichen substances were detected by TLC and MCT (Culberson & Kristinsson 1970, Culberson 1972, Orange et al. 2001).

DNA extraction, amplification, and sequencing

The dried apothecia first were checked under the dissecting microscope for well-developed fruit bodies to avoid contamination of other organisms.

Total DNA was extracted from dry apothecia following the rapid one-tube genomic DNA extraction (Steiner et al. 1995) with modifications: seven dried and cleaned apothecia were transferred directly into a 2 ml Eppendorf tube. The material was grinded with a pestle in liquid nitrogen until a fine powder was obtained. Then 150 µl TE solution was added into the tube and stirred for 2 min. until the powder was well-distributed, and immediately stored at –20°C.

Primers for PCR of the nuclear ribosomal ITS region ITS1F (Gardes & Bruns 1993) and ITS4 (White et al. 1990) were used.

The phylogenetic tree was constructed with a Bayesian approach based on the nuclear ribosomal ITS sequence data of the new species and sequences of species from the same subgenus retrieved from GenBank (Table 1).

Taxonomy

Caloplaca tianshanensis Xahidin, A. Abbas & J.C. Wei, **sp. nov.** (Figs 1, 2)

MycoBank MB 518332

Species nova similis C. peliophyllae *a qua thallo flavido-brunneo areolato cum rimis conspicuis et areolis peltatis, stipitatis in centro thalli, discis apotheciorum atris raro atrobrunneis, substantias lichenium ignotas continente differt.*

TYPE: China, Xinjiang, Mt. Nan-shan in Tianshan mountain chain, Miaoergou, on limestone, alt. 1280 m, April 10, 2009, A. Abbas & H. Xahidin 20090001 (**holotype** in XJU, **isotype** in HMAS–L).

Etymology: The specific epithet refers to the type locality.

Thallus crustose, 2–11 cm in diam., consisting of numerous peltate areoles of 0.7–3 mm wide and 0.4–0.6 mm thick, much thicker in central part of the thallus, yellowish brown, flat, separated by conspicuous cracks (Fig. 1a, b), with a whitish gray to light gray and very thin prothallus.

Upper cortex well developed, paraplectenchymatous, 50–175 µm thick; algal layer discontinuous (Fig. 1c).

Ascomata apothecia, orbicular to irregular in shape, immersed or somewhat prominent, 0.8–1 mm in diam., numerous, usually 1 per areole, sometimes 2 or occasionally more than 2, zeorine, with both a proper and a thalline margin; thalline margin raised and proper margin not visible when younger;

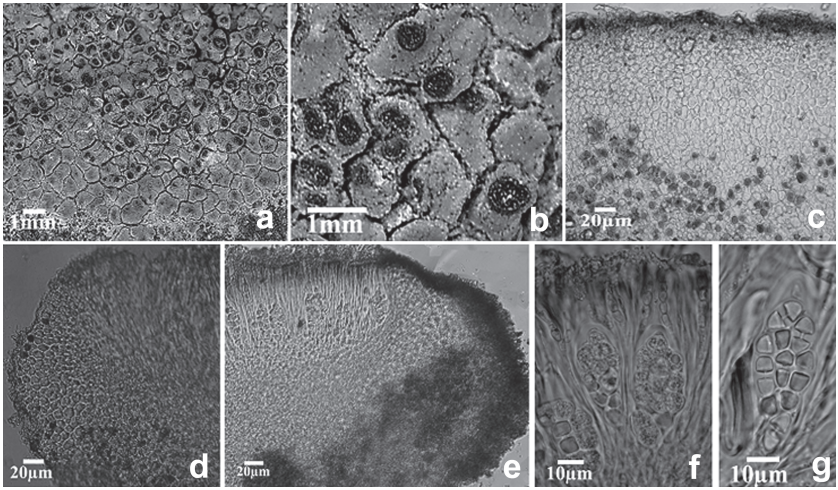

Fig.1. *Caloplaca tianshanensis*: a, b. habit; c. cross section of a peltate areole of the thallus showing the well-developed paraplectenchyma in the upper cortex; d. cross section of an apothecium showing the well-developed paraplectenchyma in the proper exciple; e. cross section of an apothecium showing the double or zeorine margin, with both thalline and proper exciple; f. cross section of the hymenium showing asci containing spores and paraphyses with beaded apices consisting of 2–5 swollen terminal cells; g. an ascus containing 8 spores with thin septa.

proper margin raised and prominent, and thalline margin lower when mature (Fig. 1e); disc dark brown to black, concave, shiny, without or with thin whitish pruina (Fig. 1a, b); hymenium 75–115 μm thick; paraphyses septate, simple, with beaded apices consisting of 2–5 swollen cells (Fig. 1f); asci 44–62 × 12–26 μm, 8–spored; spores broadly ellipsoid, polarilocular, 12–18 × 5–9 μm (Fig. 1f, g); proper exciple paraplectenchymatous (Fig. 1d); hypothecium with gray crystals, 55–90 μm thick.

Conidiomata not seen.

Chemistry: upper cortex K–, C–, epihymenium K–; two unknown substances were detected by TLC: one gives a spot in R_f class 5–6 by solvent systems A, B and G, and in R_f class 6 by solvent system C, grey-brown after charring; the other gives a spot in R_f class 5 by solvent systems A and G, in R_f class 2 by B, and in R_f class 2–3 by C, green after charring.

Remarks: The new species is similar to *C. peliophylla* in its yellowish brown thallus, but different by the areolate thallus, dark brown to black apothecium discs, the presence of two unknown lichen substances, and the Asian distribution. The latter species differs in its subsquamose thallus with shiny brown apothecia, an American distribution and the absence of lichen substances (Wetmore 1994). In addition, the new species is similar to *C. transcaspica* in its crustose and

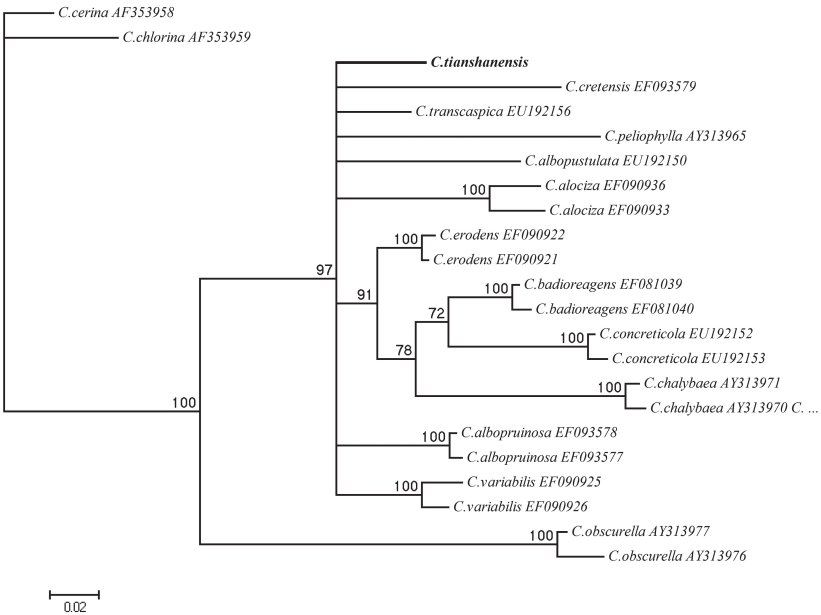

Fig. 2. Consensus tree generated by Bayesian analysis based on ITS region rDNA sequence data. *Caloplaca tianshanensis* groups with species of subgenus *Pyrenodesmia*. Bootstrap support values from 1000 replicates higher than 50% are reported at the nodes. *C. cerina* and C. *chlorina* from subgenus *Caloplaca* were selected as outgroup.

areolate thallus, but differs by its yellowish brown color, dark brown to black discs, smaller ascospores, wider isthmus in cells, and 2–5 swelling terminal cells of the paraphyses.

The ITS sequence of *C. tianshanensis* grouped with those of other 12 related species was retrieved from GenBank as a group belonging to the subgenus *Pyrenodesmia* with 100% bootstrap support. The ITS sequence of the new species *C. tianshanensis* form a distinct clade among the other 11 well recognized related species, such as *C. cretensis*, *C. transcaspica, C. peliophylla*, *C. albopustulata*, etc., with 97% bootstrap support. These results show that *C. tianshanensis* is clearly distinct from the above-mentioned well-recognized species (Fig. 2).

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